Artificial Diets for Mass Rearing The Corn Earworm (*Heliothis zea*)

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Artificial Diets for Mass Rearing the Corn Earworm (*Heliothis zea*)

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Summary

Pinto bean, CSM, WSB, and opaque-2 corn diets were tested for their suitability in mass rearing the corn earworm, *Heliothis zea* (Boddie), in the laboratory. Overall, the WSB diet was best. Although the bean diet gave more consistent results, as measured by viable adults, egg production, and egg hatch, and was nearly equal to WSB in projected insect population growth, the bean diet costs about twice as much. The CSM diet rated third. The data on the first two generations of corn earworms indicated that the opaque-2 corn diet is probably nutritionally deficient.

Introduction

Biological control of an injurious species of insect requires large numbers of laboratory-reared insects. Easily formulated and economically produced diets that rear insects competitive with those in nature are required for the success of biological control programs. The corn earworm, *Heliothis zea* (Boddie), a major pest of corn and cotton, is one insect for which biological methods of control are being studied. This paper reports information obtained from evaluation of three existing diets and one trial diet which may be used for mass rearing corn earworms.

The CSM diet,[1] the modified pinto bean diet,[2] and the WSB

[1] Burton, R. L. A low-cost artificial diet for the corn earworm. Jour. Econ. Ent. 63: 1969–1970. 1972.

[2] Burton, R. L. Mass rearing the corn earworm in the laboratory. U.S. Dept. Agr., Agr. Res. Serv. ARS–134, 8 pp. 1969.

diet[3] have shown potential for mass rearing the corn earworm. Another diet utilizing opaque-2 corn[4] as its base has produced favorable results in preliminary tests (unpublished). The constituents of these diets are readily available, economical, and easily formulated because of the small number of ingredients. The ingredients of the diets are given in table 1. In formulating the diets, the dry ingredients are blended with the water and formaldehyde in blender. The agar solution is prepared by heating the agar-water mixture to 100° C. and cooling to 60° C. before blending it with other ingredients.

[3] Burton, R. L., and Perkins, W. D. WSB, a new laboratory diet for corn earworm and fall armyworm. Jour. Econ. Ent. 65: 385–386. 1972.

[4] Seed of opaque corn was supplied by L. F. Bauman, Department of Agronomy, Purdue University, Lafayette, Ind.

TABLE 1.—*Ingredients for 1 liter of diet*

Ingredient	Diet			
	Opaque-2 corn	WSB [1]	Pinto beans	CSM [1]
Major ingredient g	135	198	111	198
Torula yeast g	33.8	10.6	33.8	10.6
Wheat germ g	---	---	52.8	---
Ascorbic acid g	3.4	3.4	3.4	3.4
Methyl *p*-hydroxy-benzoate g	2.1	2.1	2.1	2.1
Sorbic acid g	1.1	1.1	1.1	1.1
Formaldehyde, 10 percent ml	8.4	2.1	8.4	2.1
Water in blender ml	343	491	343	491
Agar g	13.5	13.5	13.5	13.5
Water for agar solution ml	338	338	338	338

[1] WSB and CSM premixes are available commercially from various companies, including Krause Milling Co., 4222 West Burnham St., Milwaukee, Wis. 53246; Lanhoff Grain Co., Danville, Ill. 61832; and Archer Daniels Midland Co., 4666 Faries Parkway, Decatur, Ill. 62521. The specifications are as follows:

CSM

Ingredient	*Percent*
Corn meal, gelatinized	63.8
Soy flour, defatted	24.2
Nonfat dry milk	5.0
Soy oil, refined	5.0
Vitamin mineral premix	2.0

WSB

Ingredient	*Percent*
Wheat fractions	73.4
Soy flour	20.0
Soy oil, stabilized	4.0
Mineral premix	2.6
Vitamin premix	0.1

Materials and Methods

The four diets—modified pinto bean, CSM, WSB, and opaque-2 corn—were evaluated in 10 trials over 4 weeks. This time allowed an adequate sample of our insect colony, which has a life cycle of about 28 days. Each trial consisted of eight replications of 25 cups of each diet, giving a total of 2,000 cups per diet for evaluation during the F_1 generation.

Each cup was charged with 10 milliliters of diet from a prepared batch of 2,500 milliliters. The cups were placed in groups of 25 in a Cel-Pack tray, infested with 1-day-old, processed *H. zea* eggs,[5] and capped. Four groups of 25 cups were placed in a bundle (four replications) and held in an environmental room at 84° to 85° F. and 55 to 60 percent relative humidity for the duration of the egg, larval, and pupal stages. The biological criteria considered in evaluating the diets were days to pupation, days to emergence, percentage viable adults, fecundity, percentage egg hatch, adult longevity, mating, and type of sperm complements.

Forty cages per diet were used to evaluate adult performance, that is, four cages containing six pairs of adults from each of the 10 trials. The adults were held at 78° to 80° F. and 75 to 80 percent relative humidity and were fed, ad libitum, a beer solution with ascorbic acid.

The eggs were collected daily on oviposition cloths. Dead moths and locked pairs were recorded and removed. Daily observations of the moths continued until all moths had died. Egg counts were made until oviposition decreased below 50 eggs per cage per day. A template, identical in size to the oviposition area but with 50 uniformly distributed circles accounting for 18 percent of the total oviposition area, was used to obtain an estimation of total oviposition. The eggs in the circles were counted and multiplied by 5.58 (the ratio of total eggs to eggs in the circles) to obtain estimates of the total number of eggs per cage. Eggs laid on the second, fourth, and sixth days were processed and used for infesting diets for the F_2 studies. Egg hatch data were obtained from eggs collected on the third and fifth days of oviposition. Four eggs were placed on the diet in each of 25 cups of each oviposition cage, and the hatch was checked 4 days later by observing the number of live larvae per cup.

Types of sperm complements were determined with two cages containing five pairs of moths per diet per trial, representing a total of 20 cages per diet. These insects were held under the same conditions as for oviposition, except the females were dissected and

[5] Processed by the method of Burton, cited in footnote 2.

checked for sperm type after two nights' exposure to the males. The sperm were typed as normal motile, normal nonmotile, mostly apyrene, or mostly eupyrene.

Second-generation studies were conducted in the same manner except that only two oviposition cages per diet were set up for each of 10 trials to obtain data on egg production. Tests were carried through the third generation on all diets except the opaque-2 corn diet. Results obtained from the first two generations on that diet suggest that additional studies be carried out to make the opaque-2 corn diet more comparable to the other diets used in the test.

All data were subjected to analysis of variance, and Duncan's multiple-range test was used to separate the means. The 0.05 level of probability was used in all analyses.

Results and Discussion

Table 2 summarizes the experimental data. Although CSM rated highest in more categories than any other diet, the differences among the diets were significant in only two categories for the entire test. In the first generation, the CSM diet resulted in a significantly shorter time to emergence than the bean and WSB diets. The WSB and bean diets were both significantly better than CSM and corn diets in producing more viable adults. WSB was significantly better than any other diet in giving greater egg production. Although egg production by WSB- and bean-fed insects was higher than that of CSM-fed insects, the number of egg-producing days remained about the same for the three diets (not shown in table 2). CSM gave a higher percentage egg hatch, but the differences among diets are not significant for this variable. The CSM and WSB diets were equivalent in producing moths capable of mating, as measured by spermatophores per female. In this respect the CSM diet was superior to the bean diet, but there was no significant difference between the mating of moths reared on the WSB and the bean diets, as measured by spermatophore count. Insects reared on the WSB diet lived an average of 1 day longer than those fed CSM and 1.4 days longer than those fed the bean diet.

In the second generation, CSM resulted in a significantly shorter time to pupation than any other diet. The corn diet produced insects that required a significantly longer time to emergence. Egg hatch was significantly higher for the WSB and bean diets than for CSM. There were no significant differences among diets in egg production in the third generation.

TABLE 2.—*Competitiveness of corn earworms reared on four artificial diets*[1]

Diet	Days to pupation	Days to emergence	Viable adults per replication of 25 cups	Egg production per female	Percentage egg hatch	Adult longevity in days	Spermatophores per female	Locked pairs per cage
			1ST GENERATION					
Opaque-2 corn	15.24a	26.26c	20.26a	778a	55.12a	12.53a	2.00a	1.50b
WSB	14.36b	25.66b	21.56b	939c	58.67a	14.53c	2.37bc	.90a
Pinto bean	14.24b	25.45b	21.76b	868b	59.55a	13.09ab	2.15ab	.82a
CSM	14.13b	25.11a	20.58a	800a	60.08a	13.57b	2.51c	1.12a
			2D GENERATION					
Opaque-2 corn	15.61c	27.71a	20.38a	614a	50.40a			
WSB	14.81b	26.24b	21.90a	757a	61.85b			
Pinto bean	14.98b	26.34b	22.56a	751a	62.90b			
CSM	14.59a	25.79b	21.30a	795a	53.10a			
			3D GENERATION					
WSB				643a				
Pinto bean				755a				
CSM				687a				

[1] Means not sharing a common letter are significantly different at the 0.05 level.

Table 3 summarizes data obtained from viable sperm studies. There were slight differences among diets in the type of sperm complements passed, but the differences were not significant.

Data from this test were used to project population growth for the various diets. The WSB and bean diets showed the greatest potential as insect diets (table 4). The WSB diet appears to be the best diet when the number of insects produced is used as an indicator. Since mating, longevity, and locking are reflected in the data on insect production, it seems appropriate to place considerable weight on the number of insects produced. Time to pupation and time to emergence are important factors in mass rearing, but differences among diets in this experiment do not seem to merit special attention to these variables.

The bean diet gave more consistent results. The average coefficient of variation (in viable adults, egg production, and egg hatch) for the bean diet was 12.7, whereas those for the WSB, CSM, and corn diets were 16.0, 16.9, and 24.3, respectively. The fact that population growth on the bean diet was 98 percent of those grown on WSB, coupled with its low coefficient of variation, favors the bean diet. However, when cost is considered, the WSB diet rates above the bean diet. The cost for 1 liter of bean diet is 25 cents, whereas the WSB and CSM diets cost about 14 cents per liter.

TABLE 3.—*Effect of diet on types of sperm complements passed during a 48-hour mating period*[1]

[Moths per cage]

Diet	Normal sperm			Mostly apyrene	No
	Motile	Nonmotile	Total	sperm	sperm [2]
1ST GENERATION [3]					
Opaque-2 corn	1.70	0.40	2.10a	0.65a	2.25
WSB	2.20	.45	2.65a	.45a	1.90
Pinto bean	2.20	.50	2.70a	.30a	2.00
CSM	2.05	.40	2.45a	.85a	1.70
2D GENERATION [3]					
Opaque-2 corn	2.31	0.0	2.31a	0.82a	1.87
WSB	2.44	.0	2.44a	.44a	2.12
Pinto bean	2.31	.19	2.50a	.76a	1.74
CSM	2.44	.19	2.63a	.68a	1.69

[1] Figures based on 5 females per cage.

[2] Also includes moths having sperm in various other categories.

[3] Means followed by the same letter are not significant at the 0.05 level.

TABLE 4.—*Projected population growth of corn earworms reared on four diets through two generations, starting with 25 larvae*[1]

Diet	Viable adults	F_1 eggs	F_1 larvae	F_1 viable adults	F_2 eggs	F_2 larvae	Percent of WSB-reared population
Opaque-2 corn	20.26	7,881	4,344	3,540	1,086,780	547,737	45
WSB	21.56	10,122	5,939	5,203	1,969,336	1,218,034	100
Pinto bean	21.76	9,444	5,624	5,073	1,904,912	1,198,190	98
CSM	20.58	8,232	4,946	4,214	1,675,065	889,460	73

[1] Assume egg hatch and egg production reflect mating data. Assume 50 percent females.

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